PB90-100850

Quantitative Analysis of Alkyl Phosphates Using Automated Cool On-Column Aqueous Injection

(U.S.) Environmental Research Lab., Athens, GA

1989

U.S. Department of Commerce
National Technical Information Service
NTIS

TECHNICAL REPORT DATA
(Please read Instructions on the reverse before completing)

1. REPORT NO.	2.	3.
EPA/600/J-89/093		PB90-100850

4. TITLE AND SUBTITLE	5. REPORT DATE
QUANTITATIVE ANALYSIS OF ALKYL PHOSPHATES USING AUTOMATED COOL ON-COLUMN AQUEOUS INJECTION	6. PERFORMING ORGANIZATION CODE

7. AUTHOR(S)	8. PERFORMING ORGANIZATION REPORT NO.
J. Jackson Ellington* and Cheryl D. Trusty**	

9. PERFORMING ORGANIZATION NAME AND ADDRESS	10. PROGRAM ELEMENT NO.
*Environmental Research Laboratory, U.S. Environmental Protection Agency, Athens GA 30613 **Technology Applications Inc., U.S. Environmental Protection Agency, Atehns GA 30613	ABWD1A 11. CONTRACT/GRANT NO.

12. SPONSORING AGENCY NAME AND ADDRESS	13. TYPE OF REPORT AND PERIOD COVERED
Environmental Research Laboratory - Athens, GA Office of Research and Development U.S. Environmental Protection Agency Athens GA 30613	14. SPONSORING AGENCY CODE EPA/600/01

15. SUPPLEMENTARY NOTES

Journal of High Resolution Chromatography 12(7):470-473, July 1989

16. ABSTRACT

A gas chromatographic method of analysis for trimethyl and triethyl phosphates was developed that used direct aqueous injection. Automated, cool, on-column injection was investigated because of the large number of samples to be analyzed and the realization that the best inlet system for quantitative high resolution gas chromatography is the non-vaporizing, cool, on-colum technique. Automated injection would increase not only sample throughput, but also precision and accuracy. Anticipated problems in the method development were the buildup of salts in the column system from injection of the buffered samples and the loss of flame in the flame ionization detector during the passage of water through the instrument.

→ con't on pg 470

17. KEY WORDS AND DOCUMENT ANALYSIS

a. DESCRIPTORS	b. IDENTIFIERS/OPEN ENDED TERMS	c. COSATI Field/Group

REPRODUCED BY
U.S. DEPARTMENT OF COMMERCE
NATIONAL TECHNICAL INFORMATION SERVICE
SPRINGFIELD, VA. 22161

18. DISTRIBUTION STATEMENT	19. SECURITY CLASS *(This Report)*	21. NO. OF PAGES
RELEASE TO PUBLIC	UNCLASSIFIED	6
	20. SECURITY CLASS *(This page)* UNCLASSIFIED	22. PRICE

EPA/600/J-89/093

Quantitative Analysis of Alkyl Phosphates Using Automated Cool On-Column Aqueous Injection

J. J. Ellington*
Environmental Research Laboratory, U.S. Environmental Protection Agency, Athens, GA 30613-7799, USA

C. D. Trusty
Technology Applications Inc., U. S. Environmental Protection Agency, Athens, GA 30613-7799, USA

Key Words:

Capillary gas chromatography
Direct aqueous automated cool on-column injection
Quantitation
Trimethyl phosphate
Triethyl phosphate

1 Introduction

The widespread use of trialkyl/aryl phosphate esters as flame retardants, plasticizers, and pesticides has resulted in the release of these potential pollutants into the environment [1-3]. The trimethyl (TMP) and triethyl (TEP) phosphates are often contaminants or impurities carried over from the synthesis of more complex phosphates [4]. TEP has been identified in the groundwater adjacent to a landfill site [5].

In ongoing research to predict the fate of chemicals released into the environment, hydrolysis rate constants of selected chemicals are experimentally determined for use in simulation models. Hydrolysis rate constant determinations require monitoring the disappearance over time of the selected chemical, at the ppm-ppb concentration range (10^{-4} to 10^{-5} M), from buffered aqueous solutions of known pH. The most direct method is quantitative analysis of the chemical in an aliquot of the reaction mixture, preferably after a chromatographic process has separated the chemical from interferences. Because of the higher water solubilities of TMP and TEP, the usual methods of isolating trialkyl/aryl phosphates from water by solvent partioning [6, 7] or sorption to a macroreticular resin [8] are not viable. *Le Bel et al.* [8] reported TEP recoveries from water of only 25 % using Amberlite XAD-2 macroreticular resin.

The objective of this research was to develop a gas chromatographic (GC) method of analysis for TMP and TEP that used direct aqueous injection. Automated, cool, on-column injection was investigated because of the large number of samples to be analyzed and the realization that the best inlet system for quantitative high resolution gas chromatography is the non-vaporizing, cool, on-column technique [9-11]. Automated injection would increase not only sample throughput, but also precision and accuracy [11]. Anticipated problems in the method development were the buildup of salts in the column system from the injection of the buffered samples and the loss of flame in the flame ionization detector during the passage of water through the instrument [12].

2 Experimental

2.1 Sample Preparation

TMP and TEP samples were prepared by spiking buffered solutions at pHs 3 (phthalate), 7 (phosphate), and 11 (phosphate) with a concentrated methanol stock to yield a maximum concentration of 35 ppm TMP in 0.025 M buffer and 24 ppm TEP in 0.0125 M buffer. Prior to injection, the pH 3 and pH 11 buffers were neutralized by the addition of seven drops of 0.5 M NaH_2PO_4 per 10 ml of buffer.

(Copyright (c) 1989 Dr. Alfred Huethig Publishers

NTIS is authorized to reproduce and sell this report. Permission for further reproduction must be obtained from the copyright owner.

2.2 Apparatus

A Hewlett-Packard 5890 GC equipped with an HP 7673A autosampler, a flame ionization detector, a cool on-column injector and an HP 3392A integrator were used for all analyses.

2.3 Gas Chromatographic Conditions

The flame ionization detector temperature was 250°C and the normal injection volume was 1.0 µl. The column temperature was held initially at 90°C for 1 min, then programmed to 115°C at 5°/min, then to 160°C at 70°/min and held at 160°C for 2 min. This temperature program was used for both columns (Section 2.4) unless noted in the figure captions. Column compensation was used. Carrier gas flow was 10 to 12 ml/min of helium with 22 to 25 ml/min of nitrogen makeup flow. Flow rates for hydrogen and air were 32 and 330 ml/min, respectively. Loss of the flame after injection of aqueous solutions was not a problem when these flow rates were maintained.

2.4 Columns and Connectors

A 1.0 to 1.5-m section of uncoated, deactivated fused silica capillary column (0.53-mm-i.d.) was used as a retention gap column. Initially, a 15-m x 0.53-mm-i.d. DB-5 column having a film thickness of 1.0 µm (J&W Scientific) was used in the analyses. A metal Valco fitting was used to connect the 1.5-m retention gap column (Alltech Associates) to the analytical column. This system was used for both compounds over a 75-day period for more than 500 injections. A 13-m x 0.53-mm DB-Wax column having a film thickness of 1.0 µm (J&W Scientific) connected to a 1.0-m, uncoated, deactivated 0.53-mm-i.d. retention gap column with an all-glass pressfit connector (Hewlett-Packard) also was used.

2.5 Column Rinsing

Columns (with connected retention gaps) were removed from the GC and backflushed with sequential, 10-ml rinses of water, methanol, acetonitrile, and isooctane. The rinse connection was made to the detector end of the column. After the last solvent rinse, the column system was dried with a stream of nitrogen, reinstalled in the GC, and conditioned by a 6°/min temperature program from 60°C to the upper limit of the GC temperature program used for analysis.

3 Results and Discussion

3.1 Precision of Injections and Calibration

After establishing that the flame would not be extinguished by 1-µl buffered aqueous injections, calibration standards of TMP and TEP were prepared and injected using the autosampler. **Table 1** gives the analytical precision details from multiple injections of the standards. As expected, the percent relative standard deviations were larger at lower concentrations than at the higher concentrations. Detector response was linear in the concentration ranges investigated and the lower limit of detection was in the ppb range. The percent relative standard deviation for manual injections ranged from 1% at the higher concentrations to 5% at the lower concentrations.

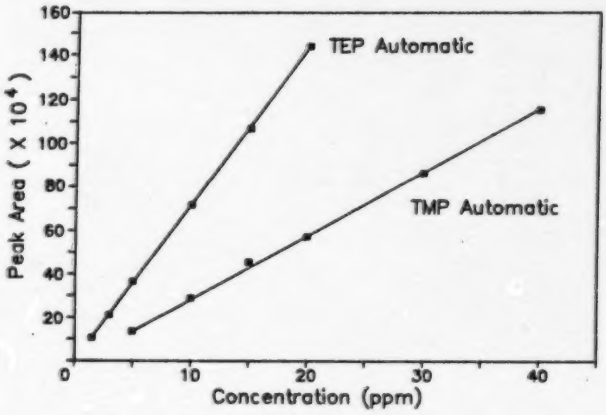

Figure 1

Calibration line for automatic injection of TMP and TEP on the DB-Wax column.

Table 1

Precision of multiple autoinjections on the DB-Wax Column[a]

Compound	Conc. (ppm)	*n*	Mean area ($\times 10^{-6}$)[a]	% RSD
TMP	40	5	1.12	0.17
	30	5	0.84	0.42
	20	5	0.58	0.52
	15	5	0.44	0.38
	10	5	0.30	0.40
	5	5	0.15	0.72
TEP	20	4	1.30	0.58
	15	4	1.03	0.38
	10	4	0.68	1.82
	5	5	0.33	3.12
	3	4	0.20	3.35
	1.5	5	0.10	2.47

[a] The mean of the area values were multiplied by 10^{-6} and rounded.

To establish linearity of detector response, duplicate, 1-µl injections were made to permit correlation of peak area counts (detector response) with concentration (**Figure 1**) in the 5 to 40 ppm concentration range for TMP and the 1.5 to 20 ppm concentration range for TEP. The response *versus* concentration data were processed using a data entry/linear regression program (Lotus 1-2-3).

Figure 1 is a plot of the linear regression lines for the automated calibration injections of TMP and TEP on the DB-Wax column using the data in Table 1. Similar plots were obtained for the remaining automated and manual calibration runs. The correlation coefficient values (r^2) were > 0.997. Thus, both methods of injection yield excellent fit to the regression line.

Tables 2 and **3** contain values for the slope, the percent standard error of the slope, and the *y*-intercept for manual and automated injections of TMP and TEP on DB-5 and DB-Wax columns. Automated injections in each case yielded a steeper

regression line, smaller standard error of the slope, and closer intercept to the origin. The greater relative deviation of area counts for manual injections, particularly at the lower concentrations, is the most probable cause of devation from the ideal calibration line that intercepts the origin.

Table 2

Calibration regression data for TMP.

Inj. tech/column	Slope ($\times 10^{-4}$)[a]	Std. error of slope as % of slope	*y*-intercept ($\times 10^{-4}$)[a]
Manual/DB-5	N/A[b]		
Automated/DB-5	2.9[c]	1.4	0.0
Manual/DB-Wax	2.6[c]	1.6	2.2
Automated/DB-Wax	2.8[d]	1.0	1.6

[a] Values for slope and *y*-intercept were multiplied by 10^{-4} and rounded.
[b] Not available, corresponding data not generated on 1-µm film thickness column.
[c] Average of area counts from duplicate injections were used in regression analyses.
[d] Data from Tabel 1 were used in regression analysis.

Table 3

Calibration regression data for TEP.

Inj. tech/column	Slope ($\times 10^{-4}$)[a]	Std. error of slope as % of slope	*y*-intercept ($\times 10^{-4}$)[a]
Manual/DB-5	6.0[b]	1.6	−2.2
Automated/DB-5	7.2[b]	0.5	−0.1
Manual/DB-Wax	5.9[b]	2.6	−1.8
Automated/DB-Wax	6.6[c]	2.1	0.6

[a] Values for slope and *y*-intercept were multiplied by 10^{-4} and rounded.
[b] Average of area counts from duplicate injections were used in regression analysis.
[c] Data from Table 1 were used in regression analysis.

Figure 2

Close-up photograph (50x) of salt buildup in retention gap taken with an Olympus BHTU Biological Microscope.

3.2 Accumulation of Buffer Salts

Approximately 0.4 mg of buffer salts were injected onto the retention gap/analytical column per 100 injections. Most of this salt was retained on the retention gap column, but some moved through the analytical column and was deposited in the column and in the detector housing. This salt buildup (**Figure 2**) was

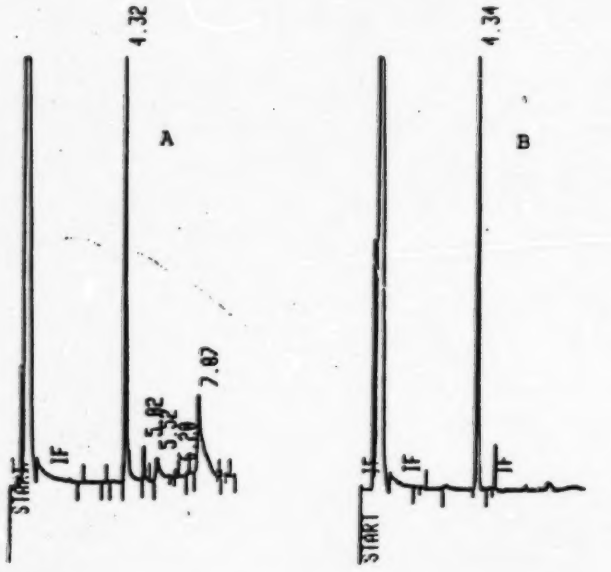

Figure 3

Chromatograms of TMP (4.32 min) on the DB-Wax column before rinsing (A) and after rinsing (B). Temperature program was 95°C for 1 minute then 10° per minute to 160°C.

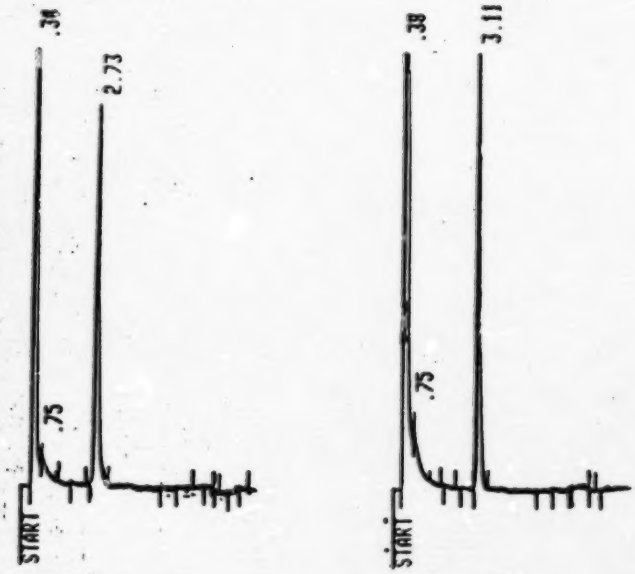

Figure 4

Chromatograms of TMP (2.73 min) and TEP (3.11 min) on the 1 µm film thickness DB-5 Column.

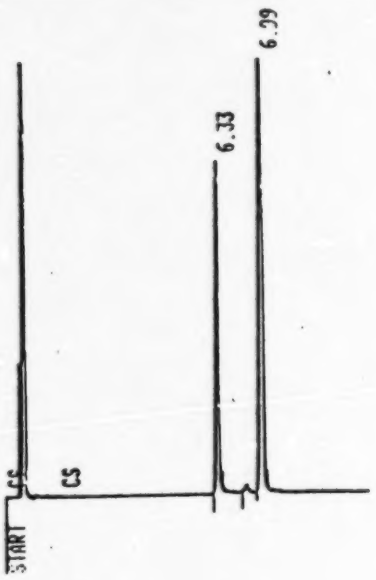

Figure 5

Chromatogram of TMP (6.33 min) and TEP (6.99 min) on the 1 μm film thickness DB-Wax column.

easily visible with a 20X magnifier. Periodic cleaning of the detector and housing as well as rinsing of the retention gap/analytical column was required. **Figure 3** demonstrates the benefits of column rinsing; chromatogram A is a TMP sample before the retention gap/analytical column was rinsed (after salt buildup) and chromatogram B is the same sample after the retention gap/analytical column was rinsed.

The noisy baseline and the appearance of the unknown peak at 7.87 min in Figure 3A signaled the need for rinsing and temperature conditioning. The columns typically gave the responses illustrated in **Figures 4** and **5**.

When the DB-5 column used for the analyses in Figure 4 (1-μm film thickness) could no longer be regenerated by solvent rinsing, attemps to analyze TMP on another DB-5 column with a 1.5-μm film thickness were unsuccessful. The TMP peak consistently split, whereas the TEP peak shape was normal. The most likely explanation of the peak splitting is that a fraction of the TMP chromatographed and coeluted with the water while the remaining fraction was retarded by the liquid phase and eluted as the trailing peak. Although retention times for TMP and TEP under identical flow and temperature program conditions are longer on the DB-Wax column (Figures 4 and 5) peak splitting was not a problem because water was apparently absorbed by the stationary phase and eluted after TEP [12].

4 Conclusions

A quantitative method for analyzing water-soluble alkyl phosphates using automated, cool, on-column aqueous injections has been developed. The buildup of buffer salts is a problem that can be dealt with by routinely rinsing the column with solvents and periodically replacing the retention gap column. Periodic cleaning of the detector and detector housing also is recommended. Hydrogen and air flows were established that maintained the hydrogen flame during the elution of water.

References

[1] *S. S. Lande, J. Santodonato, P. Howard, D. Greninger, D. Christopher,* and *J. Saxena,* "Investigation of Selected Potential Environmental Contaminants: Haloalkylphosphates", U.S. Environmental Protection Agency, Washington, DC, EPA-560/2-76-007 (1976).

[2] *T. W. Lapp,* "The Manufacture and Use of Selected Aryl and Alkyl Phosphate Esters", U.S. Environmental Protection Agency, Washington, DC, EPA 560/6-76-008 (1976).

[3] *N. N. Melnikov,* Chemistry of Pesticides, Springer-Verlag, New York (1971).

[4] *M. Marcus, J. Spigarelli,* and *H. Miller,* "Organic Compounds in Organophosphorus Pesticide Manufacturing Wastewaters", U.S. Environmental Protection Agency, Athens, GA, EPA-600/4-78-056 (1978).

[5] *W. J. Dunlap, D. C. Shew, M. R. Scalf, R. L. Cosby,* and *J. M. Robertson,* In "Identification and Analysis of Organic Pollutants in Water", *L. H. Keith,* ed., Ann Arbor Science, Ann Arbor, Michigan (1976) 453-477.

[6] *M. T. Shafik* and *H. F. Enos,* J. Agr. Food Chem. **17** (1969) 1186.

[7] *M. T. Shafik, D. Bradway,* and *H. F. Enos,* J. Agr. Food Chem. **19** (1971) 885.

[8] *G. L. Le Bel, D. T. Williams,* and *F. M. Benoit,* J. Assoc. Off. Anal. Chem. **64** (1981) 991.

[9] *K. Grob, Jr.* and *H. P. Neukom,* J. Chromatogr. **189** (1980) 109.

[10] *J. V. Hinshaw,* J. Chromatogr. Sci. **26** (1988) 142.

[11] *D. Kennedy,* HRC & CC **11** (1988) 350.

[12] *B. S. Middleditch, Nak-June Sung,* and *A. Zlatkis,* Chromatographia **23** (1987) 273.

Note: Mention of trade names or commercial products does not constitute endorsement or recommendation for use by the U.S. Environmental Protection Agency.

Ms received: January 21, 1989